Exhibit III

Influence of Immunological Factors in Respiratory Syncytial Virus Disease

Of the Lower Respiratory Tract

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DURING THE past few years there has developed a growing suspicion that immunological factors play a major role in the most severe type of pulmonary damage associated with respiratory syncytial (RS) infection.[1,2] This view has evolved from a consideration of several unusual aspects of the epidemiological pattern of RS virus disease of early life and more recently from attempts to interpret the altered reactivity to RS virus infection exhibited by recipients of an inactivated RS vaccine. In these two instances, one an experiment of nature and the other of man, it appears that the presence of virus in the respiratory tract is not sufficient to explain the severe disease manifestations of the young infant or the altered reactivity of vaccinees. It is clear that the affected host contributes certain factors to the equation of disease which are as essential as the virus itself.

In this presentation we will describe those properties of the virus and its ecological pattern which bear upon the question of host immunological participation in naturally occurring RS virus disease of early infancy. The phenomenon of vaccine-induced altered reactivity and its relevance to the pathogenesis of natural disease will also be discussed. Finally, consideration will be given to a beneficial type of immune response which appears to provide resistance to infection and to act in opposition to those immune mechanisms which contribute to development of disease.

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The Virus and Its Ecological Patterns

Respiratory syncytial virus is a medium sized (120 mμ to 200 mμ) enveloped virus which contains lipid and RNA and which matures at the limiting membrane of the infected cell.[3] Its internal component exhibits helical symmetry while its outer envelope is studded with spike-like projections.[4,5] Although there is some controversy concerning the dimensions of the inner helix, it appears to have a diameter of 13 mμ.[4,5] It is thus intermediate in size between the helix of the influenza viruses (9 mμ) and that of the paramyxoviruses (18 mμ). For this reason, it has been suggested that RS virus and pneumonia virus of mice, which also has a 13 mμ inner helix, may comprise a third subgroup of myxoviruses distinct from the influenza myxoviruses and the paramyxoviruses.[5]

Our acquaintance with RS virus dates back 13 years to the time when we recovered the first human strains from children with lower respiratory tract disease.[6] During the ensuing interval, a consistent pattern of the epidemiology of RS virus infection has emerged from studies performed in many different geographic areas. It is now a well-established fact that this virus is the most important respiratory tract pathogen of early life.[7-9] Severe disease manifestations occur most commonly in young infants. In addition, infection, which usually represents reinfection, occurs commonly in older children and adults, but the associated disease is generally mild. In large urban population

Table 1.—*Age Distribution of RS Virus Illness in Vaccine Group Compared With That of Community (Children's Hospital of Washington, DC, 1966 to 1967)**

Age at Which RS Bronchiolitis or Pneumonia Occurred (Mo)	Patients From Community			RS Vaccinees (No.)	
	No.	Percent of Total			
1-2	36	31	(65%)	3	
3-4	21	18			
5-6	18	16		4	
7-8	13	11		2	(63%)
9-10	5	4		1	
11-12	4	3		3	
13-14	4	3		3	
15-16	5	4		2	
17-18	2	2		1	
19-20	3	3			
21-22	4	3			
23-24	1	1			
Total	116			19	

* Modified from Kim et al, 1969.[10]

centers, RS virus produces an extensive epidemic every year which is easily recognized by the dramatic increase in hospitalization of young infants for serious lower respiratory tract disease.[8,6]

Within the past few years, it has become clear that the ecological pattern of RS virus infection in young infants is unusual in several respects. Probably the most remarkable feature of this virus is the age distribution of patients with severe lower respiratory tract illness. Serious RS virus disease occurs most often in young infants in the first few months of life, and the frequency of disease then decreases with increasing age.[1,2,8] This pattern is not seen with any of the other recognized respiratory tract pathogens.[8] Representative data obtained during the 1966 to 1967 period in the Washington, DC, area are shown in Table 1. Serious RS virus disease occurred most commonly in infants 1 to 2 months of age, and 65% of patients with RS respiratory tract illness requiring admission to the hospital were in the first half year of life.[10]

The age-illness relationship just described resembles the age distribution of passively acquired serum-neutralizing antibody.[2] All adults thus far tested possess moderate to high levels of serum-neutralizing antibody, and this is transferred quantitatively from the mother to the infant. It has been shown, with the plaque-reduction technique, that infants possess passively acquired serum-neutralizing antibody until the sixth to seventh month of life.[2] The level of such passive antibody is generally inversely related to age and decreases approximately twofold each month.

Infants who develop RS virus disease do not differ from their non-ill cohorts with respect to serum antibody. Acute phase sera of young infants admitted to the hospital with RS virus bronchiolitis or pneumonia contain moderate to high levels of serum-neutralizing antibody, as measured by the plaque-reduction technique.[1,2] The age distribution of antibody titers is similar to that of normal infants in that the level of antibody is inversely related to age. However, the mean antibody levels of infants with RS disease are approximately twofold lower. This difference could occur because of prior RS infection in a few of the normal infants included in our survey or possibly it could result from the development of antigen-antibody complexes in the affected lung tissue of infants with RS disease, leading to depletion of serum antibody.

Relation of Serum Antibody to Resistance

The behavior of RS virus in young infants provides an almost unique opportunity to assess the protective effect of serum antibody for this agent in the absence of any potential effect of local respiratory tract antibody. In older individuals, this type of opportunity is rarely encountered with RS or other respiratory viruses because infection generally induces the development of both local respiratory tract and serum antibodies. Thus, individuals who have undergone infection often possess both types of antibodies, and there is a rough correlation in the levels of these substances.[11,12]

As mentioned earlier, the young infant possesses maternally-derived serum antibody at the time RS infection occurs whereas local antibody, which is not transferred passively from the mother, is presumably not present. In this setting, the observation that serious RS disease occurs most often in infants who possess the highest levels of passively transferred antibody clearly indicates that serum-neutralizing antibody (pre-

Table 2.—*Relation of Preexisting Serum-Neutralizing Antibody to Pneumonia and Infection in Nonvaccinated Residents of Nursery During RS Virus Outbreak December 1966 to January 1967**

Reciprocal of Serum Dilution Which Produced 60% Reduction of RS Virus Plaque†	Total Population Studied by Virus-Recovery Technique			Group Studied by Both Virus Recovery and CF Antibody Techniques	
	No. of Nursery Residents‡	No. Who Developed Pneumonia	No. From Whom Virus Recovered	No. of Nursery Residents	No. With Evidence of RS Virus Infection, ie, Virus Recovery or CF Antibody Rise or Both
<20	28	3 (11%)	21 (75%)	20	20 (100%)
20-400	21	3 (14%)	9 (43%)	10	9 (90%)
401-1280 or >	36	5 (14%)	6 (17%)	21	12 (57%)
Total	85	11 (13%)	36 (42%)	51	41 (80%)

* Modified from Kapikian et al, 1969.[13]
† Tested with RS virus strain from outbreak, using sera collected just prior to outbreak.
‡ Includes only individuals in residence for at least one week during outbreak period, Dec 4, 1966, to Jan 31, 1967.

sumably IgG) per se does not provide effective protection against the most serious effects of RS virus. The frequency with which the virus infects young infants also indicates that serum antibody does not provide effective resistance to infection.

Additional evidence that serum-neutralizing antibody does not provide effective protection was obtained during a recent study of an RS virus outbreak which occurred in a closed nursery, whose residents ranged in age from 6 to 65 months.[18] Preoutbreak serum specimens were available from 85 of the nursery residents, and these specimens were assayed for neutralizing antibody by the plaque-reduction technique. As seen in Table 2, preexisting serum-neutralizing antibody did not appear to confer resistance to pneumonia in the population under study. It should be emphasized that the pneumonic illnesses which developed in the older infants and young children were not severe.

Relation of Local Antibody to Resistance to RS Infection

Older children and adults develop less serious illness during RS virus reinfection than do young infants undergoing primary infection. Since serum-neutralizing antibody does not appear to be protective, it seemed to us that local respiratory tract, secretory, 11S IgA antibody might act as the main determinant of resistance and be responsible for the milder illnesses associated with reinfection. To clarify this situation, a series of studies were performed to evaluate the properties and effects of local antibody. Initially, antibody in nasal secretions was characterized and shown to be predominantly 11S IgA (J. Mills, MD, unpublished data). Each of 104 adults tested possessed such RS-specific secretory antibody, and the levels varied within a wide range. In addition, experimentally infected adults and naturally infected infants and children were shown to develop neutralizing activity in their nasal secretions during convalescence.[12,14] The latter finding indicated that the respiratory tract secretory antibody system operates early in life.

To test the hypothesis that local respiratory tract antibody was the main determinant of resistance to RS virus, we tested the effect of this antibody upon experimental RS virus infection in adult volunteers.[12] Sixteen volunteers were selected for nasopharyngeal challenge, using 500 plaque-forming units (pfu) of virus. Eight men had a low level of nasal-secretion neutralizing activity, eight had a high level, and there was no overlap in level of antibody between the two groups (Table 3). Each of the volunteers possessed a moderately high level of serum-neutralizing antibody. Following challenge, each of the men in the two groups shed virus, and the temporal pattern of virus shedding was the same for men in either group. However, when the virus content of the nasopharyngeal washings was estimated by the plaque technique, it was found that the low nasal antibody group shed large quantities of virus (up to 10^5 pfu per ml of nasopharyngeal washing) whereas the washings of the high antibody group contained very little virus (Table 3). This difference was related to nasal antibody and not serum

Table 3.—*Influence of Nasal Secretory IgA Upon Response of Volunteers to Experimental Challenge With 500 PFU of RS Virus**

Group	Volunteer	Prechallenge Nasal Antibody Titer† (Reciprocal)	Response to Experimental Challenge: 4 Fold or Greater Rise in Serum-Neutralizing Antibody	Nasal Washings Neutralizing Antibody	Serum CF Antibody	Any Immunological Response	Peak Virus Titer of Throat Washings, LOG_{10} PFU‡ Per ML
Low nasal antibody§	1	10	+	+	+	+	5.0
	2	7	+	+	+	+	4.1
	3	8	0	+	+	+	1.8
	4	10	0	+	0	+	2.5
	5	18	0	+	0	+	3.2
	6	8	0	0	+	+	1.8
	7	11	0	0	0	0	0.9
	8	11	0	0	0	0	<0.3
High nasal antibody‖	9-16	87 to 775 (geometric mean = 248)	0	0	0	0	<0.3 to 0.3

* Modified from Mills et al, 1969.[12] † Adjusted to 20 mg IgA per 100 milliliters.
‡ Determined in Hep_2 petri dish cultures; antibody titers determined in same system and calculated for 60% plaque-reduction endpoint by probit analysis.
§ Serum-neutralizing antibody titers, prechallenge, ranged from 1:321 to 1:1700; geometric mean = 1:890.
‖ Serum-neutralizing antibody titers, prechallenge, ranged from 1:590 to 1:10,000; geometric mean = 1:2300.

antibody since the two groups differed primarily in their level of nasal antibody.

Following infection, none of the high-nasal antibody men developed serum-neutralizing, serum complement-fixing (CF) or nasal-secretion neutralizing antibody. In contrast, six of the eight low-nasal antibody group developed a rise in one or more of these antibodies (Table 3). The findings from this study indicate that local antibody did not influence susceptibility to infection with 500 pfu of RS virus; however, the level of local antibody had a marked effect upon the level of virus replication and, secondarily, upon the immune response to infection.[12] This interpretation supports the thesis that resistance to RS virus disease is a function of local respiratory tract secretory antibody.

Altered Reactivity to RS Virus Infection Following Parenteral Administration of Inactivated Vaccine

The most recent experience which bears upon the potential role of immunological factors in RS virus disease was acquired during a series of controlled field evaluations of an inactivated RS vaccine. This vaccine was prepared in vervet monkey kidney tissue culture and contained virus which had been concentrated 100 fold and adsorbed to alum. The vaccine appeared to be antigenic, even when given to infants and young children who lacked detectable serum-neutralizing antibody. In one study, each of seven seronegative subjects (serum-neutralizing antibody titer <1:20 by the highly sensitive plaque-reduction technique) who received two or three doses of vaccine developed serum-neutralizing antibody titers of 1:1000 to 1:10,000 or greater.[13] In addition, 18 of 19 vaccinees with prevaccination serum-neutralizing antibody titers of 1:25 to 1:250 developed four to 400 fold rises in antibody levels after one to three doses of vaccine. Similarly, in another study, 91% of young vaccinees developed complement-fixing antibody after one to three injections of vaccine.[10]

Additional evidence for antigenicity of the vaccine is presented in Table 4 which contrasts the levels of serum-neutralizing antibody of infants and children who had received RS vaccine with those of unvaccinated controls. These serum samples were obtained from residents of a nursery just prior to an extensive outbreak of RS virus infection, which involved all seronegative children (Table 2). At this time, which represented from one to nine months after administration of the first dose of vaccine, each of the 29 vac-

Table 4.—*Serum Neutralizing Antibody Status of Vaccinated and Nonvaccinated Nursery Residents Just Prior to Onset of RS Virus Outbreak**

Reciprocal of Serum Dilution Which Produced 60% Reduction of RS Virus Plaques†	No. of Nursery Residents of Indicated Age in Months‡									
	Vaccinees					Nonvaccinated Group				
	6-11	12-23	24-35	36-58	Total	6-11§	12-23	24-35	36-65	Total
<20					0	4 (57%)	19 (53%)	4 (15%)	1 (6%)	28 (33%)
20-100					0	2	5	3	1	11 (13%)
101-400		2	1	1	4 (14%)		3	5	2	10 (12%)
401-1,280	1		2	1	4 (14%)	1	5	8	5	19 (22%)
>1,280		6	6	9	21 (72%)		4	6	7	17 (20%)
Total	1	8	9	11	29	7	36	26	16	85

* Modified from Kapikian et al, 1969.[13]
† Tested with RS virus strain recovered from nursery outbreak.
‡ Includes only individuals in residence at least seven days during outbreak period, Dec 4, 1966 to Jan 31, 1967.
§ Includes one child aged 5 months.

cinees tested possessed serum-neutralizing antibody at a titer of 1:101 to 1:1280 or greater whereas 46% of the nonvaccinated control group lacked detectable antibody (33%) or had a level <1:100 (13%) (Table 4). This difference, which was particularly striking in the 12 to 23 month age group, undoubtedly reflected an antigenic effect of the vaccine. Parenthetically, the proportion seronegative non-vaccinated nursery residents decreased from 57% in the 6 to 11 month age group to 6% in the 36 to 65 month group, indicating that the plaque-reduction technique for assay of antibody was specific, in addition to being quite sensitive.

When an RS virus outbreak occurred in the nursery approximately nine months after initiation of the vaccine trial, the frequency with which virus was recovered from vaccinees was not significantly different from that of the unvaccinated control group (Table 5). This indicated that vaccine-induced serum antibody, although present at moderately high levels, did not protect against infection. More striking than the failure of vaccine to protect against the virus was the unexpected response to infection exhibited by the vaccinees. Nine of 15 (60%) vaccinees 6 to 23 months of age developed pneumonia or bronchiolitis or both, in five instances serious enough to require hospitalization, whereas only four of 47 (8%) unvaccinated controls developed pneumonia and none of these children required admission to the hospital.[18] This difference in response indicated that the vaccine had induced an altered, exaggerated reactivity to infection. Definite evidence of such an altered response to infection was not seen in the older vaccinees, 24 to 65 months of age (Table 5).

In another study, involving infants who were seen as outpatients at a child health center, evidence of vaccine-induced altered reactivity to RS virus infection was again seen in an unmistakable manner.[10] When RS virus was prevalent during the winters of 1965 to 1966 and 1966 to 1967, the frequency of infection among RS vaccinees (74%) was not remarkably different from that of age-matched subjects who received either a tissue culture-grown type 1 parainfluenza vaccine or an egg-grown trivalent parainfluenza vaccine (53%) (Table 6). However, at the time of their RS infection, 78% of the RS vaccinees required hospitalization because of serious obstructive lower respiratory tract disease whereas only 5% of the infected parainfluenza vaccinees developed illness of similar severity. The illnesses experienced by the RS vaccinees were more severe than those seen in age-matched unvaccinated patients from the community. The parainfluenza type 1 vaccine used in this study was prepared in vervet monkey kidney tissue culture in a manner virtually identical to that of the RS vaccine; in fact, the two vaccines were prepared by the same pharmaceutical manufacturer. Since the parainfluenza type 1 vaccinees did not exhibit an exaggerated response to RS virus infection, it is unlikely that monkey kidney antigens

Table 5.—*Frequency of Pneumonia and RS Virus Isolation by Age and Vaccine Status in Residents of Two Nursery Cottages**

Age (mo)	Vaccinated Group: No. in Group	Vaccinated Group: No. With Pneumonia	Vaccinated Group: No. From Whom RS Virus Isolated	Unvaccinated Group: No. in Group	Unvaccinated Group: No. With Pneumonia	Unvaccinated Group: No. From Whom RS Virus Isolated
6-11	2	2	1	7	1	4
12-23	13	7 } 9† (60%)‡	9 } 10 (67%)	41	3 } 4 (8%)‡	19 } 23 (48%)
24-35	11	4	3	30	6	15
36-65	11	0 } 4 (18%)	5 } 8 (36%)	31	1 } 7 (11%)	4 } 19 (31%)
Total	37	13 (35%)	18 (49%)	109	11 (10%)	42 (39%)

* Modified from Kapikian et al, 1969.[13]
† Five of these children were seriously ill and required hospitalization.
‡ Percentages significantly different ($P < 0.0001$).

Table 6.—*RS Virus Infection and Illness in Groups of Infants Who Received One or More Injections of Inactivated RS or Parainfluenza Virus Vaccine**

Vaccine: Host System	Vaccine: Virus	Vaccine: Concentration, Adjuvant	No. of Vaccinees	No. of Vaccinees Who Underwent Natural Infection With RS Virus Subsequent to Vaccination†	No. of Vaccinees Who Developed Illness at Time of RS Infection: Bronchiolitis or Pneumonia	No. of Vaccinees Who Developed Illness at Time of RS Infection: Bronchitis or URI	No. of Vaccinees Requiring Hospitalization at Time of RS Illness
Vervet monkey kidney tissue culture	RS	100X, alum	31	23	19	4	18 (78%)
	Type 1 parainfluenza	100X, alum	20 } 40	12 } 21	2 } 4	10 } 17	1 } 1 (5.0%)
Egg	Types 1, 2, and 3 parainfluenza	3X, no adjuvant	20	9	2	7	0

* Modified from Kim et al, 1969.[10]
† Based upon virus recovery or CF antibody rise or both.

were responsible for the altered reactivity associated with the RS vaccine. Instead, it appears that a component of the RS virus itself was responsible for the RS vaccine effect.

Pathogenesis of Naturally Occurring Disease and Vaccine-Induced Altered Reactivity

Several observations, which were cited previously, indicated that serum-neutralizing antibody per se did not provide effective protection against RS virus lower respiratory tract disease. When viewed in another manner, these same observations provide a basis for speculating that serum antibody may actually contribute to the development of serious lung damage during infection in infancy.[1,2] Thus, RS virus bronchiolitis and pneumonia occur most commonly during the first few months of life, which is the time when maternally transmitted antibody is present and at its highest level. Subsequently, both the incidence of disease and the level of passively acquired antibody regress in a similar manner with increasing age. In this sense, passively transmitted antibody and RS virus disease behave in a parallel fashion with respect to age.

Any hypothesis which attempts to explain the pathogenesis of serious RS disease of early life must take into account the intrinsic pathogenic potential of the virus itself.

virus, in the nursery outbreak described above, as well as during a previous RS outbreak in this nursery, the virus caused pneumonia in children who lacked detectable serum antibody (Table 2).[13,15] If immunological factors are important in lower respiratory tract disease, they act to enhance the basic pathogenic effects of the virus. We propose that the interaction of serum antibody and viral antigens in the lungs plays an important role in producing the disease manifestations which are characteristic of RS illness of early infancy, especially the bronchiolar obstruction of bronchiolitis. If this supposition is correct, then local respiratory tract antibody and serum antibody act in opposition to each other. Local secretory antibody, as described earlier, appears to prevent infection or moderate its extent while, in our view, serum antibody probably is an active participant in the process of tissue damage.

At this point, although one can only speculate upon the basis for immunological enhancement of RS virus disease of infancy, several possibilities can be excluded with reasonable assurance. Type 1 (anaphylactic) and type 4 (cell-mediated delayed hypersensitivity) immune reactions are not involved since neither reaginic antibody (IgE) nor cell-mediated sensitivity are transferred from mother to infant.[16] IgA and IgM antibodies are also not transferred across the placenta and for this reason could not participate in the postulated pulmonary RS antigen-antibody reaction.[16] This leaves maternally transmitted serum IgG antibody as the probable immunological component of the disease complex. Whether such serum IgG reacts with virus antigen on the infected cell surface to produce a type 2 immune (cytolytic) reaction or reacts with soluble antigens of the virus in an extracellular location to effect a type 3 immune reaction is a matter for conjecture.

Since complement is often fixed during type 2 and type 3 immune reactions, we recently tested ten infants who were in the acute phase of RS bronchiolitis for evidence of complement consumption (P. Sta Ana, et al, unpublished data). These young patients were found to have normal serum levels of both total complement and C/3. In addition, using the immunofluorescence technique, it was not possible to detect fixation of complement or C/3 in the affected lungs of three infants who died from RS virus disease (P. Ward et al, unpublished data). Either complement plays no role in the postulated serum antibody-RS antigen immunological reaction or the quantity of complement fixed during the reaction is insufficient to deplete the serum pool or be detected by immunofluorescence.

Of course, it is possible that an age-related nonimmunologically determined factor(s) could be responsible for the unusual pattern of RS virus illness in early infancy. This would mean that the age-disease-antibody relationships which exist are merely fortuitous. The simplest nonimmunological explanation for the unusual predilection for RS virus disease in early infancy is that the air pathways at this time are smaller and more easily obstructed than later in life. This anatomic factor may make a contribution to the occurrence of RS disease. At this point, it should be emphasized that the majority of RS vaccinees who developed severe obstructive pulmonary disease were older than the usual unvaccinated infant with bronchiolitis.[10,13] As shown in Table 1, 65% of nonvaccine-associated RS virus disease in the community occurred in infants in the first half year of life whereas 63% of the vaccinees who exhibited altered reactivity to RS virus developed obstructive disease after 6 months of age. The development of severe, typical, RS-obstructive bronchiolitic disease by the older vaccinees makes it unlikely that the size of the air pathway is a major determinant in RS illness.

There are several mechanisms whereby RS antigen, given parenterally, could induce an altered reactivity to subsequent RS infection. At this time, we favor a unitary hypothesis which links this phenomenon to the increased reactivity to virus seen during unmodified infection of early infancy.[2] In both conditions, disease probably results from the interaction of serum antibody and RS antigen in the lungs of individuals who lack respiratory tract secretory antibody or who possess a level of this antibody which is insufficient to confer protection. Several observations provide support for this unitary hypothesis. The clinical picture and course of vaccine-altered disease was remarkably

similar to that of the naturally occurring illness of young infants. Furthermore, in the nursery study, vaccine-altered reactivity to RS infection was limited primarily to the younger vaccinees (Table 5).[18] These vaccinees, however, were beyond the age at which severe RS lower respiratory disease usually occurs. The younger vaccinees had the least prior experience with RS infection (Table 4), and, presumably, they possessed the least amount of local respiratory antibody. By stimulating serum antibody in these individuals with inactivated vaccine, an imbalance in serum and respiratory tract antibodies was created. This type of imbalance also occurs in the young infants who possess maternally transmitted serum antibody but, presumably, not respiratory secretory antibody.

At present, the possibility that delayed hypersensitivity played a major role in vaccine-induced altered reactivity to RS virus infection cannot be ruled out. However, as indicated previously, it is extremely unlikely that this mechanism is involved in the naturally occurring disease of young infants.

Recently, evidence for delayed hypersensitivity to RS virus was sought, using the lymphocyte transformation technique which is thought to measure cell-mediated reactivity (S. Leiken et al, unpublished data). Skin testing was not possible since appropriate reagents were not available. The lymphocytes of infants who had undergone natural infection with RS virus transformed when exposed to a suspension of concentrated RS virus-infected monkey kidney tissue culture material whereas this did not occur with uninfected monkey kidney culture antigens. Similarly, lymphocytes from RS vaccinees who had not been infected with RS virus also transformed in the presence of RS antigens. Two adults with moderately high levels of serum-neutralizing antibody were also tested, and their lymphocytes were transformed by RS antigens. The extent of transformation was approximately two times greater for RS vaccinees, whether or not infection had occurred, than for unvaccinated infants or children infected under natural conditions. These findings suggest that both RS infection and vaccination with an inactivated vaccine induced delayed hypersensitivity to RS antigens and that vaccination was the more effective stimulus (S. Leiken et al, unpublished data).

Although many, and perhaps most, older children and adults would be expected to possess lymphocytes sensitized to RS viral antigens, such individuals develop less serious RS respiratory tract disease than do infants. This suggests that delayed hypersensitivity per se does not necessarily lead to the development of lower respiratory tract disease during RS infection. If delayed hypersensitivity does play a role in RS immunopathology, it is likely to be of importance only when respiratory tract secretory antibody is not sufficient to prevent extensive replication of the virus. This situation may have occurred in the infants who received inactivated RS vaccine; however, the pathogenic significance of delayed hypersensitivity cannot be inferred since serum antibody was also induced by the vaccine.

While it remains to be proved that RS virus disease of infancy contains a strong immunopathological component, it might be worthwhile to speculate upon the factors which would be of importance if such is the case. An understanding of the constellation of factors which may coalesce to produce frequent serious RS disease might help explain why other respiratory tract pathogens fail to produce a similar disease pattern. For RS virus to succeed in producing an immunologically determined illness in young infants, four conditions are probably essential, and these constitute the tetrad of RS virus disease of early infancy. These essential factors include: (1) a virus which produces specific antigen at the affected cell surface and an excess of soluble viral antigens during infection of the lower respiratory tract; (2) absence of protective local respiratory secretory antibody; (3) presence of maternally derived antibody in serum through the sixth to seventh month of life; and (4) a high rate of infection during the first half year of life when passively acquired antibody is present in serum.

Many respiratory tract agents are probably disqualified from producing "RS-like" disease by their failure to meet condition number 4. Influenza A virus may be unable to produce an immunologically determined illness during early infancy because maternally derived serum antibody does not corre-

d, in most instances, to the antigenic akeup of the current epidemic strain. The nly respiratory tract pathogen, other than S virus, which meets the four conditions escribed above is type 3 parainfluenza vi-us. With this agent, we do not observe the ramatic occurrence of illness in the first ew months of life followed by a decreasing ncidence with increasing age. However, sev-ral years ago we did find that lower re-piratory tract disease caused by type 3 arainfluenza virus occurred as often under months of age as after 6 months whereas he other respiratory tract pathogens (other han RS virus) appeared to produce serious lisease less frequently during early infancy.[8]

Relevance of Immunopathological Factors in RS Disease to Development of Effective Vaccines

The observations and speculations pres-nted have relevance to current efforts to levelop a safe effective vaccine for preven-ion of serious RS disease in early life. If, as s now suspected, an immunological reaction n[illegible]ing serum antibodies plays an impor-[illegible]ole in pathogenesis of RS disease, [illegible]hasis should be given to methods which ure effective in stimulating local respiratory ract secretory antibody. This might be ichieved by the nasopharyngeal instillation of either inactivated antigen or live atten-uated virus. It is also quite clear that RS virus serum antibodies should not be stimu-lated without concomitant induction of re-spiratory tract antibodies lest an imbalance be created which could lead to a state of altered reactivity during subsequent infec-tion.

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